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14. ABSTRACT
Our objective in this effort was to determine the source(s) of tripton absorption and how this absorption affects the optical properties of seawater. Tripton is non-phytoplankton particulate matter. In work completed, no humic fluorescence signature has been observed in the particulate fraction. Additionally, a relatively high refractive index has been estimated for tripton, consistent with the hypothesis that a significant portion of tripton absorption may be derived from inorganic minerals.Results thus far have implications for predicting radiative transfer in marine waters.

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Quantification and Characterization of the Absorption by Non-Phytoplankton Particulate Material

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LONG-TERM GOALS

Our long-term goals are to better understand the absorption and scattering properties of the constituents of seawater, the temporal and spatial variability in these properties, and how these properties alter the submarine and water-leaving light fields. Of particular interest in this effort are the absorption properties of non-phytoplankton particulate material, also known as tripton. Although evidence suggests tripton absorption is a ubiquitous component of total absorption and attenuation in marine waters, the primary absorbing substance(s) are not known. Thus, predicting the temporal and spatial variability of tripton and its associated optical properties is difficult. Characterizing the optical properties and composition of tripton will fill a critical gap in our understanding of the factors influencing radiative transfer in marine waters.

OBJECTIVES

Our overall objective is to determine the source(s) of tripton absorption and how this absorption affects the optical properties of seawater.

Since tripton is a diverse assemblage of materials (organic and inorganic, alive and dead, small and large, new and old), the corresponding chromophores are expected to be similarly diverse. One of our first order objectives is, thus, to identify the dominant chromophore groups and to characterize the optical properties of each group. We intend to develop novel experimental approaches, involving fluorescence spectroscopy and specially tailored chemical and physical extraction techniques, to accomplish this task.

A second order objective of this work is to determine how the absorbing constituents of

tripton affect the overall optical properties of tripton. For example, the absorption coefficient and the spectral character of tripton will be different if the absorbing constituent typically coats particles (adsorption of humic material, e.g.) or if it is evenly dispersed within particles (inorganic minerals such as quartz, e.g.). This is the 'packaging effect.' Scattering efficiencies for these particles will also be different, because absorption will reduce the scattering efficiency of the particle, thus reducing the bulk scattering coefficient of the mixture. Once the primary chromophore groups are identified, we then intend to address these issues.

APPROACH

Our level 1 approach is to use a combination of extraction techniques (Figure 1) coupled with absorption and fluorescence measurements to determine the absorption properties of the predominant individual chromophore groups of tripton. Each of the following chromophore groups in tripton are being investigated: 1) humic material associated with particles, 2) residual phytoplankton pigments associated with particles, 3) chromophoric cellular material such as cytochromes, and 4) inorganic minerals.

The chromophores of tripton are being identified and characterized using multi-excitation fluorescence spectroscopy (ISA-SPEX Fluorolog-3-21) and spectrophotometry with a reflecting tube absorption meter (ac-9). Protocols for using the ac-9 reflective tube absorption meter have been established previously (Twardowski et al. 1999). Protocol development for using the Fluorolog-3 for our specific applications has been delegated to Dr. Russ Desiderio (OSU). When possible, absorption coefficients will be obtained from fluorescence by converting emission to absorption based on 'relative fluorescence efficiencies' (emitted photons/absorbed photons) determined in the laboratory and from the literature for specific chromophore groups.

Samples for laboratory analyses have been collected in tandem with vertical profiles of hydrographic and optical parameters in Newport, OR (a day trip in March 2000) and

Potential visible light-absorbing constituents of particulate material →
Potential visible light-absorbing constituents of tripton →

treatment ↓	sorbed humic material	residual phytoplankton pigments	chromophoric cellular material (cytochromes)	inorganic minerals	phytoplankton cellular pigments
methanol extraction	?	X	10% or less		X
base extraction	X	?			
combustion	X	X	X		X
altering pH	X	?			
exposure to UV	X	X	X		X
lysing with lysozyme			X		X

Figure 1. Methodological approaches to chemically and physically isolating individual chromophores of particulate material.

Puget Sound, WA (2 week cruise at the end of March 2000). These samples generally had high levels of tripton, dominated in some cases by organic detritus and in others by inorganic minerals. Efforts thus far have concentrated on testing the hypothesis that humic material in the particulate fraction is the primary absorbing component of tripton.

The relationship between the distribution of an absorbing substance within a particle and the bulk optical properties of a particle population are being investigated using theoretical models. Boss and Twardowski (submitted) and Twardowski et al. (submitted) have addressed the effects of an evenly dispersed absorbing substance throughout a particle on spectral attenuation and backscattering ratios, respectively. E. Boss will also use a coated sphere Mie theory model (Bohren and Huffman 1983) to assess the effects of a particle coated with an absorbing substance. This model may be applicable when substances such as humic materials adsorb onto the surface of particles.

WORK COMPLETED

This report addresses work completed primarily in the period from January 2000 to May 2000. Since May, the PI has changed institutions and participated in a 10 week Navy-ASEE Summer Fellowship.

After procuring and installing the Fluorolog-3 spectrofluorometer, the instrument was characterized by Dr. Russ Desiderio. Dr. Desiderio has prepared a detailed protocols document for the instrument. Based on a series of tests, the document describes the necessary corrections for our applications, including corrections for order sorting by the monochromators, spectral signal and reference corrections, quantum corrections, dark count corrections, and blank subtraction. The accompanying software can perform some of these corrections, but, in some cases, Dr. Desiderio has determined that corrections made by the software were inaccurate. These inaccuracies have been verified by ISA-SPEX senior spectroscopists. Moreover, the impressive thoroughness of Dr. Desiderio's characterization of the instrument gives us a great deal of confidence that our quantum-corrected spectra are as accurate as possible.

The Fluorolog-3 has been moved to the PI's new institution and is currently being installed.

In the samples collected from Newport, OR and Puget Sound, WA, I have concentrated on using UV excitation to stimulate fluorescence by humic substances in whole and filtered samples. I have also made these measurements with the 'front face' accessory which allows the fluorescence of particles to be directly measured off a filter pad while minimizing contamination from reflection of the excitation beam. The in-situ hydrographic and optical data from the Puget Sound cruise has also been processed and is being analyzed.

A theoretical model for determining particle composition (refractive index) from the backscattering ratio and spectral attenuation was developed (Twardowski et al. submitted). The model has been applied to data collected from the Gulf of California in 1998 and from data collected at the Rutgers LEO site in July 2000. The model uses scattering information to determine whether tripton is predominantly comprised of organic or inorganic material.

Humic fluorescence measurements were made at the Rutgers LEO site in July 2000 with and without a 0.2 μm prefilter to assess the presence of humic materials in the particulate fraction. This data is currently being processed.

At the ONR CoBOP 2000 exercise, Boss and Zaneveld (OSU) collected humic fluorescence measurements with the WET Labs Safire with and without a 0.2 μm prefilter. This data is currently being processed.

RESULTS

In the samples from Newport and Puget Sound we have observed no detectable fluorescence signature from humic materials in the particulate fraction. This observation does not support our original hypothesis that the source of tripton absorption is humic materials associated with particulate matter through adsorption or intraparticle polymerization reactions. It is important to note that the particles were not concentrated before measurement. However, the absence of humic material in the particulate fraction may be a more representative observation than originally thought.

From the theoretical modeling efforts (Boss and Twardowski, submitted; Twardowski et al., submitted), we have determined that evenly dispersed absorption within particles will have a significant impact on the scattering properties of tripton only at relatively high levels. Applying the model to data from the Gulf of California also indicated that the tripton-dominated water below the thermocline consistently had a relatively high refractive index, about 1.010-1.012 relative to water. This observation can be explained in two ways: 1) the particles were predominantly organic, but with much lower water content than phytoplankton (like fecal pellets, perhaps), or 2) the particle population had a significant component derived from inorganic minerals.

Although the majority of the proposed experimental tests have not been completed, we believe the results thus far are compelling. The observations of 1) no humic material in the particulate fraction, and 2) a relatively high refractive index for tripton, are consistent with the hypothesis that a significant portion of tripton absorption may be derived from inorganic minerals. Typical absorption spectra from inorganic minerals (e.g., Tassan and Ferrari 1995; Ahn 1999), indeed, resemble measured spectra for tripton (monotonically decreasing with increasing wavelength). Admittedly, however, we are working with a limited data set at this point and must await the results from our other proposed experiments to amass the evidence to make definitive conclusions.

IMPACT/APPLICATIONS

The lack of a humic fluorescence signature in the particulate fraction was not expected. Although preliminary, this observation suggests that another component may be important. A likely candidate chromophore is inorganic minerals. If inorganic minerals are significant, the temporal and spatial dynamics of tripton absorption will be different than previously postulated. The apparent ubiquity of tripton absorption will not be due to an equilibrium condition between the dissolved and adsorbed particulate phases of humic

material. Instead, tripton absorption may be linked to processes such as resuspension of sediments, aeolian deposition, and/or the in-situ formation of 'hard' biological structures.

It is expected that results from this work will lead to the development of a technique for the direct in-situ determination of phytoplankton absorption, separate from that of tripton. Phytoplankton absorption is needed to model photosynthetic capacities and primary productivity in natural phytoplankton populations.

TRANSITIONS

R. Arnone (NRL Stennis) is in the process of transitioning a 'version 0' total suspended matter algorithm based on the work of Twardowski et al. (submitted) that will be implemented by his remote sensing image acquisition and processing infrastructure automatically, in near-real time.

The refractive index scattering model (Twardowski et al. submitted) and the size distribution model (Boss and Twardowski, submitted) are now being used to identify bulk particle composition in several studies, including data collected under ONR Thin Layers, ONR HyCODE, ONR/NOPP sponsored research in Puget Sound (M.J. Perry's group), and NASA SIMBIOS (e.g., Boss et al. submitted; Boss et al. 2001).

During the summer of 2000 (while Twardowski was on sabbatical), the Fluorolog-3 was used by Dr. Kelly Faulkner's group (OSU) in the development of an in-situ detection method for barium.

RELATED PROJECTS

As a result of a Navy-ASEE Fellowship, Twardowski is working with Bob Arnone (NRL Stennis), Alan Weidemann (NRL Stennis), Curt Davis (NRL Washington), and Vlad Haltrin (NRL Stennis) to develop a remote sensing algorithm for total suspended matter (Twardowski et al. 2001) based on the model of Twardowski et al. (submitted).

Paul Hill (Dalhousie) and Boss are investigating the flocculation of tripton in the bottom boundary layer using the ONR Coastal Mixing and Optics data set.

In data from the 1998 ONR Thin Layers exercise, Twardowski, Tim Cowles (OSU), and Russ Desiderio (OSU) have identified layers of humic fluorescence in the particulate fraction. In this same data set, Lisa Eisner (OSU), Cowles, and Twardowski are investigating the relationship between HPLC pigment concentrations and particulate absorption spectra (Eisner et al. 2000; in prep.). A method was developed in this study to deconvolve particulate attenuation spectra into tripton and phytoplankton components. Related work has been presented by Boss et al. (1999), Coleman et al. (2001), Dekshenieks et al. (accepted), Rines et al. (accepted), and Twardowski and Donaghay (1998; accepted; in press).

Al Hanson (SubChem, Inc. and URI) and Twardowski are developing new in-situ chemical detection methods using fluorescence. R&D will be done on the Fluorolog-3.

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